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Thrombin Generation and Activity During Thrombolysis and Concomitant Heparin Therapy in Patients With Acute Myocardial Infarction

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Objectives. **This prospective study investigated the behavior of thrombin generation and activity during thrombolysis and concomitant heparin administration.**

Background. **It has been shown that during thrombolytic therapy there is an increase in thrombin generation and activity. Increased thrombin activity is suppressed by concomitant intravenous heparin, but it is unknown whether thrombin generation is also affected.**

Methods. **Thrombin generation was assessed by measuring prothrombin fragment 1 + 2 and thrombin–antithrombin complex plasma levels and thrombin activity by measuring fibrinopeptide A plasma levels. Serial blood samples were obtained before and at 90 min and 24 and 48 h after the administration of streptokinase (15 patients), recombinant tissue-type plasminogen activator (15 patients) or anistreplase (13 patients). An intravenous bolus of heparin (5,000 IU) was administered before the start of thrombolytic therapy, followed by an infusion of 1,000 U/h to maintain an activated partial thromboplastin time >1.5 times baseline.**

Results. **During thrombolytic and concomitant heparin therapy, there was an increase in the plasma levels of prothrombin fragment 1 + 2 (baseline 1.08 vs. 2.73 nmol/liter, $p < 0.001$) and thrombin–antithrombin complex (baseline 6.5 vs. 17.1 μg/ml, $p < 0.01$) at 90 min, whereas no change was observed in fibrinopeptide A at 90 min (baseline 2.8 vs. 3.0 nmol/liter, p = NS).**

Conclusions. **During thrombolytic therapy with both fibrin-specific and non–fibrin-specific drugs, there is an increase in thrombin generation despite concomitant administration of intravenous heparin.**

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Failure to achieve reperfusion and the occurrence of subsequent reocclusion after successful thrombolysis are major limitations of thrombolytic therapy in acute myocardial infarction. Recent biochemical evidence has shown that thrombolytic agents activate both platelets (1,2) and clotting factors (3–8). The activation of the hemostatic system is considered to play a pivotal role in preventing or retarding fibrinolytic-induced coronary reperfusion and in producing early reocclusion, and this has led to the recommendation of adjunctive antithrombotic therapy with aspirin and heparin (9). The clinical benefit of the addition of aspirin to thrombolysis has already been demonstrated (10); the clinical benefit of the addition of heparin to thrombolysis is still controversial (11,12).

Thrombin plays a central role in the pathogenesis of coronary artery thrombosis and rethrombosis. It is a serine protease generated by factor Xa activity through the cleavage from prothrombin of a 32-amino acid fragment, called prothrombin fragment 1 + 2. Free thrombin is inactivated by antithrombin as soon as it is generated, and the thrombin–antithrombin complex is cleared. If the generation of thrombin is such that it overwhelms this natural anticoagulatory mechanism, the thrombin cleavage of the 16-amino acid peptide fibrinopeptide A from fibrinogen initiates fibrin formation. The availability of specific immunoassays for measuring the plasma levels of prothrombin fragment 1 + 2 (13), thrombin–antithrombin complex (14) and fibrinopeptide A (15) allows thrombin generation and activity to be evaluated under in vivo conditions. Previous studies have shown that, during thrombolysis with either recombinant tissue-type plasminogen activator (rt-PA) or streptokinase (6–8), there is an increase in the

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plasma levels of thrombin–antithrombin complex and fibrinopeptide A, and this increase can be suppressed by heparin (6). During rt-PA treatment, there is also an increase in prothrombin fragment 1 + 2 (5), but it is not yet known whether this can be prevented by heparin. Although heparin is very effective in inhibiting thrombin activity on fibrinogen (16), it seems to be less effective in inhibiting factor Xa activity on prothrombin, especially when factor Xa is incorporated in the tenase complex (17).

In the present study we prospectively measured the plasma concentrations of prothrombin fragment 1 + 2, thrombin–antithrombin complex and fibrinopeptide A in patients with acute myocardial infarction who were receiving thrombolytic therapy in conjunction with heparin. The simultaneous measurement of these markers, which monitor different steps of the coagulation cascade, allowed different levels of activation of the hemostatic mechanism to be detected under in vivo conditions.

Methods

Between April 1990 and July 1991, 115 consecutive patients <70 years old admitted to the coronary care unit of the Second Division of Cardiology, Niguarda Hospital, Milan, with a diagnosis of myocardial infarction and no contraindications to thrombolytic therapy were considered eligible to enter the study. Inclusion criteria were 1) a history of chest pain lasting at least 30 min but no longer than 6 h that failed to respond to sublingual or intravenous nitrates; 2) ST segment elevation or depression ≥0.2 mV in at least two contiguous electrocardiographic leads evolving into pathologic Q waves and ST-T wave changes; and 3) development of elevated creatinine kinase and MB fraction levels of at least twice the upper normal limit.

Exclusion criteria were 1) concomitant peripheral vascular disorders or valvular heart disease (9 patients); 2) previous coronary artery bypass surgery or coronary angioplasty (5 patients); 3) anticoagulant treatment before baseline blood sampling (40 patients); 4) comorbid conditions known to affect coagulation activation markers (malignancy, sepsis, nephritis, collagen disorders, coagulation disorders) (6 patients); and 5) difficult venous access (12 patients).

Study design. This was a prospective study of consecutive patients. After determination of eligibility and inclusion into the study, a baseline blood sample was obtained from enrolled patients before any treatment was started. Subsequently, all patients received aspirin (165 to 325 mg) and an intravenous bolus of heparin (5,000 IU), immediately followed by a continuous intravenous infusion of heparin (1,000 IU/h), which was then adjusted to maintain the activated partial thromboplastin time at >1.5 times baseline values. Thrombolytic therapy was started immediately after the administration of the heparin bolus. The patients were randomized to receive 1) rt-PA, 10 mg bolus followed by an additional 50 mg over the first hour and 40 mg over the next hour; 2) streptokinase, an infusion of 1,500,000 U over 1 h; 3) anistreplase, 30 U infused over 5 min. Additional treatments included intravenous beta-adrenergic blocking agents (5 mg of intravenous atenolol, then 50 to 100 mg/day orally [12 patients]), nitrates (0.1 to 0.8 μg/kg body weight per min for 48 h [30 patients]), angiotensin-converting enzyme inhibitors (5 to 10 mg/day of oral lisinopril [6 patients]). Additional blood samples for coagulation activation markers were obtained 90 min and 24 and 48 h after the start of thrombolytic therapy.

Blood sampling and processing. Venipunctures were performed atraumatically by three specially trained investigators (P.A.M., L.O., A.S.) using 19-gauge butterfly infusion sets and the two-syringe technique. Samples at different times were obtained by separate venipunctures, and inadequate samples were prospectively discarded. Because all of our blood samples were obtained by means of clean venipunctures and meticulous technique to minimize in vitro artifacts, the values of the coagulation activation markers are much lower than those reported in other studies (3,6) during which the blood samples were obtained using indwelling catheters.

After the first 4 ml of blood was discarded, samples were collected directly into refrigerated vacutainers containing an anticoagulant mixture composed of a thrombin inhibitor, ethylenediaminetetraacetic acid (EDTA) and aprotinin purchased from Byk-Sangtec (Dietzenbach, Germany). The ratio of anticoagulant to blood was 1:9 (vol/vol). Blood samples were immediately centrifuged at 2,500 *g* for 25 min at 4°C; the platelet-free plasmas were frozen on dry ice and stored at −80°C until analyzed.

Biochemical determinations. All samples were analyzed by investigators who were not aware of the clinical data. Plasma levels of prothrombin fragment 1 + 2 were measured using double-antibody radioimmunoassay as previously described (18). This method has an interassay coefficient of variation of ~8%. The plasma levels of thrombin–antithrombin complex were measured using a commercially available kit (Enzygnost TAT kit, Beringwerke AG, Marburg, Germany). The coefficient of variation of this method is ~6%. The plasma concentrations of fibrinopeptide A were determined in duplicate by means of enzyme immunoassay in plasma extracted twice with bentonite to remove fibrinogen (Diagnostica Stago, Asnieres, France). This technique has an interassay coefficient of variation of ~5%.

Informed consent. The study was approved by the Institutional Review Board of the Ca' Granda Niguarda Hospital, and informed consent was obtained from all subjects. All of the clinical studies and informed consent procedures were also approved by the Committee on Clinical Investigations of the Beth Israel Hospital.

Statistical analysis. The deviations from a normal distribution of plasma concentrations of prothrombin fragment 1 + 2, thrombin–antithrombin complex and fibrinopeptide A were tested by calculating the coefficients of skewness and kurtosis. Because the plasma levels of coagulation activation markers were found to be non-normally distributed, paired data were analyzed by means of the Wilcoxon signed-rank test or, for multiple comparisons, by the Friedman test. Kruskal-Wallis one-way analysis of variance was used to test the

Table 1. Clinical Characteristics of the Study Patients

	rt-PA (n = 15)	Streptokinase (n = 15)	Anistreplase (n = 13)
Age (yr)	63 ± 8	57 ± 7	58 ± 7
Male gender	12 (80)	13 (86)	12 (80)
Smokers	10 (66)	11 (73)	11 (73)
Hypertension	3 (29)	2 (13)	1 (6)
Diabetes mellitus	1 (6)	2 (13)	1 (6)
Cholesterol (mg/dl)	220 ± 24	207 ± 22	227 ± 39

Data presented are mean value ± SD or number (%) of patients. rt-PA = recombinant tissue-type plasminogen activator.

difference between groups; subsequent pairwise comparisons were made using the Mann-Whitney *U* test. Descriptive statistics include mean values and standard deviations or medians, 25th and 75th and 10th and 90th percentiles as appropriate. All presented tests are two-tailed; p values <0.05 were regarded as statistically significant. The number of patients who exhibited prothrombin fragment 1 + 2 and fibrinopeptide A plasma concentrations of above the upper normal limit was calculated by determining the 95th percentile of the distribution in a control group of 32 healthy subjects matched for age and gender with the study patients, which was set at 1.02 nmol/liter for prothrombin fragment 1 + 2, 4 μg/liter for thrombin-antithrombin complex and 2.2 nmol/liter for fibrinopeptide A.

Results

Serial blood samples were taken from 43 patients (37 men, 6 women; mean [±SD] age 58 ± 7 years). Twenty patients had an infarction of the anterior wall, 15 of the inferior wall and 8 of the posterior wall. The mean time between the onset of symptoms and the start of thrombolytic therapy was 3.5 ± 1.5 h. Fifteen patients received rt-PA, 15 patients streptokinase and 13 patients anistreplase. No significant difference was observed in clinical characteristics of the three treatment groups (Table 1).

Prothrombin fragment 1 + 2, thrombin–antithrombin complex and fibrinopeptide A plasma levels at different time points in the 43 study patients. Figure 1 shows medians and 25th, 75th, 10th and 90th percentiles of prothrombin fragment 1 + 2, thrombin–antithrombin complex and fibrinopeptide A plasma levels before and after thrombolytic therapy. There was a significant change in prothrombin fragment 1 + 2 plasma concentrations at the different time points ($p < 0.0001$). From a baseline level of 1.08 nmol/liter (interquartile range 0.71 to 1.66), prothrombin fragment 1 + 2 plasma levels increased to 2.73 nmol/liter (interquartile range 2.0 to 3.48) 90 min after the start of thrombolytic therapy ($p < 0.001$). No difference in comparison with baseline was observed after 24 h (1.03 nmol/liter, interquartile range 0.69 to 1.35, $p = 0.8$) or 48 h (1.08 nmol/liter, interquartile range 0.8 to 1.57, $p = 0.7$). Abnormal values of prothrombin fragment 1 + 2 were found in 31 (72%) of the 43 patients at baseline, 40 (93%) of 43 at 90 min, 22 (53%) of 41 at 24 h and 21 (60%) of 35 at 48 h.

There was a significant change in thrombin–antithrombin complex plasma concentrations at the different time points ($p < 0.001$). From a baseline of 6.5 μg/liter (interquartile range 5.2 to 10.3), thrombin–antithrombin complex plasma levels increased to 17.1 μg/liter (interquartile range 12.5 to 26.6) 90 min after the start of thrombolytic therapy ($p < 0.01$). No difference in comparison with baseline was observed after 24 h (7.1 μg/liter, interquartile range 3.9 to 13.3, $p = 0.6$) or 48 h (5.8 μg/liter, interquartile range 3.8 to 7.7, $p = 0.1$). Abnormal values of thrombin–antithrombin complex were found in 33 (76%) of the 43 patients at baseline, 40 (93%) of 43 at 90 min, 22 (53%) of 41 at 24 h and 16 (46%) of 35 at 48 h.

There was a significant decrease in fibrinopeptide A plasma concentrations at the different time points ($p < 0.01$). Fibrinopeptide A plasma levels were 2.8 nmol/liter (interquartile range 1.3 to 4.1) at baseline and 3.0 nmol/liter (interquartile range 1.45 to 4.5) 90 min after the start of thrombolytic therapy ($p = 0.8$). In comparison with baseline there was a trend toward a decrease in fibrinopeptide A plasma levels after 24 h (1.8 nmol/liter, interquartile range 1.3 to 2.8, $p = 0.051$) and 48 h (2.0 nmol/liter, interquartile range 1.1 to 2.5, $p = 0.019$). Abnormal values of fibrinopeptide A were found in 25 (59%) of the 43 patients at baseline, 25 (59%) of 43 at 90 min, 19 (46%) of 41 at 24 h and in 16 (46%) of 35 at 48 h.

Prothrombin fragment 1 + 2, thrombin–antithrombin complex and fibrinopeptide A plasma levels according to the thrombolytic drug administered. Figure 1 shows individual values of prothrombin fragment 1 + 2, thrombin–antithrombin complex and fibrinopeptide A plasma levels before and after thrombolytic therapy according to the thrombolytic drug administered. Median prothrombin fragment 1 + 2, thrombin–antithrombin complex and fibrinopeptide A plasma levels, as well as their interquartile ranges as functions of the thrombolytic drug administered at different times are given in Table 2. There were no significant differences in the baseline plasma levels of the coagulation activation markers among the three treatment groups. Whichever thrombolytic drug was given, in comparison with baseline, at 90 min there was a significant increase in prothrombin fragment 1 + 2 ($p < 0.01$) and thrombin–antithrombin complex plasma levels ($p < 0.01$), whereas no significant change in fibrinopeptide A plasma levels was observed. Prothrombin fragment 1 + 2 and thrombin–antithrombin complex plasma levels at the 24- and 48-h determinations were not significantly different in comparison with baseline, whereas the 48-h plasma levels of fibrinopeptide A were significantly lower than baseline values in all three treatment groups ($p < 0.05$).

No differences were detected among the three groups in terms of median changes in prothrombin fragment 1 + 2 at different times. At 90 min there was a median increase of 1.45 nmol/liter (interquartile range 0.58 to 3.02) in the rt-PA group, 1.67 nmol/liter (interquartile range 0.88 to 2.65) in the streptokinase group and 1.77 nmol/liter (interquartile range −16 to 2.79) in the anistreplase group ($p = 0.95$). Compared with baseline, at 24 h there was a median change of 0.02 nmol/liter (interquartile range −0.29 to 0.44) in the rt-PA

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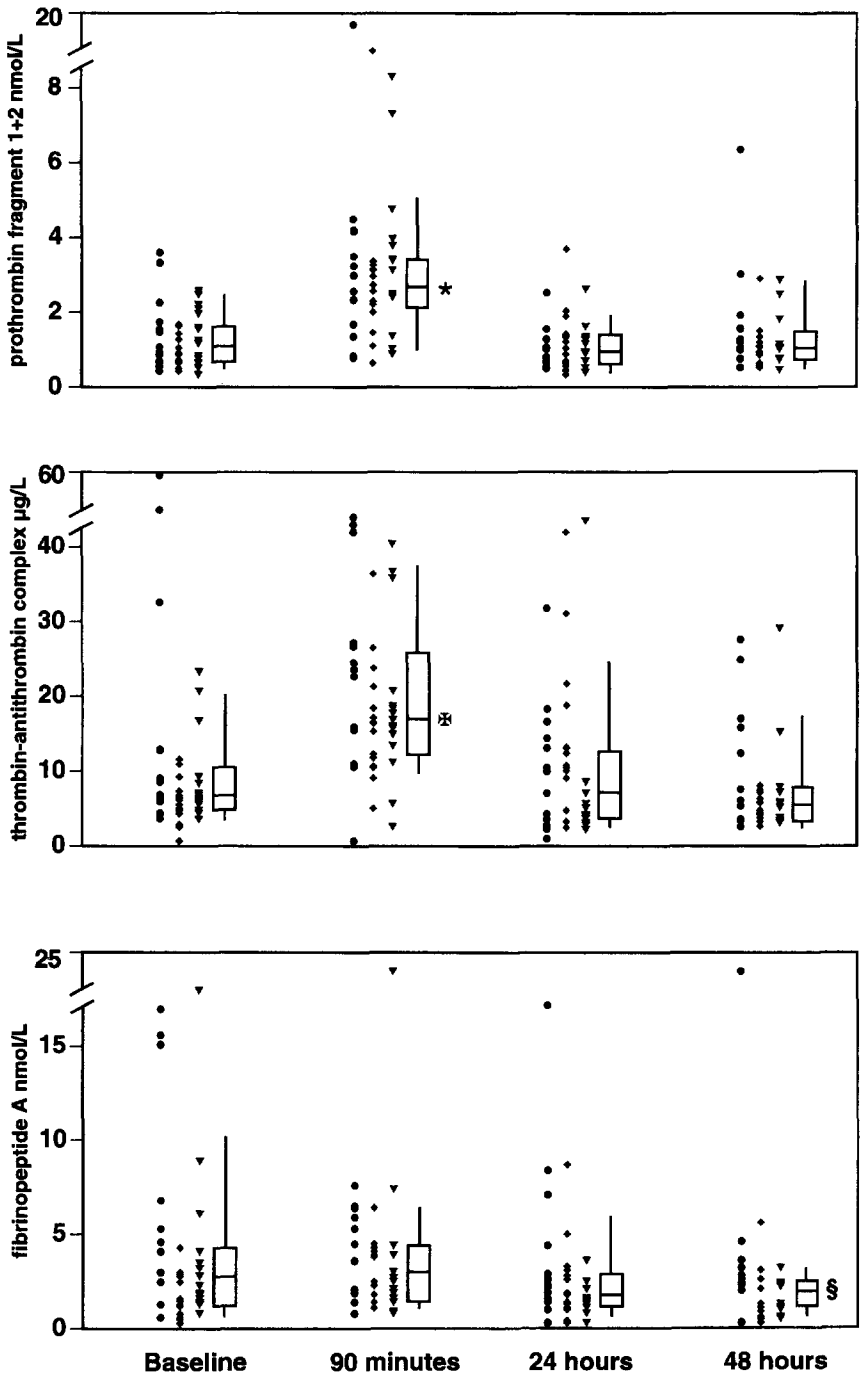

Figure 1. Plots of prothrombin fragment 1 + 2 (**top**), thrombin–antithrombin complex (**middle**) and fibrinopeptide A (**bottom**) plasma levels in 43 patients with acute myocardial infarction who received intravenous heparin in addition to thrombolytic therapy with recombinant tissue-type plasminogen activator (●), streptokinase (◆) or anistreplase (▼). Samples were obtained at hospital admission (baseline) and 90 min, 24 h and 48 h after the start of thrombolytic therapy. **Plot boxes** with median values and 25th, 75th, 10th and 90th percentiles refer to the whole study group. *$p < 0.001$, #$p < 0.01$, §$p < 0.05$ versus baseline.

group, −0.13 nmol/liter (interquartile range −0.26 to 0.75) in the streptokinase group and −0.39 nmol/liter (interquartile range −1.08 to 0.46) in the anistreplase group ($p = 0.58$). At 48 h there was a median change of 0.19 nmol/liter (interquartile range 0.17 to 0.35) in the rt-PA group, −0.08 nmol/liter (interquartile range −0.42 to −0.07) in the streptokinase group and 0.13 nmol/liter (interquartile range −0.64 to 1.01) in the anistreplase group ($p = 0.56$).

No differences were detected among the three groups in terms of median changes in thrombin–antithrombin complex at different time points. At 90 min there was a median increase of 10.1 µg/liter (interquartile range 2.4 to 14.8) in the rt-PA group, 5.6 µg/liter (interquartile range 5 to 17.9) in the streptokinase group and 9.4 µg/liter (interquartile range 0.55 to 19.2) in the anistreplase group ($p = 0.94$). Compared with baseline, at 24 h there was a median change of −0.5 µg/liter (interquartile range −4.8 to 1.8) in the rt-PA group, 2.7 µg/liter (interquartile range −5 to 13.6) in the streptokinase group and −1.6 µg/liter (interquartile range −21 to 4.45) in the anistreplase group ($p = 0.24$). At 48 h there was a median change of −1.15 µg/liter (interquartile range −2.5 to 1.5) in the rt-PA group, 1.7 µg/ml (interquartile range −6.8 to

Table 2. Plasma Concentrations of Prothrombin Fragment 1 + 2, Thrombin–Antithrombin Complex and Fibrinopeptide A According to the Different Thrombolytic Drugs Administered

	Prothrombin Fragment 1 + 2 (nmol/liter)			Thrombin–Antithrombin Complex (μg/liter)			Fibrinopeptide A (nmol/liter)		
	rt-PA (n = 15)	Streptokinase (n = 15)	Anistreplase (n = 13)	rt-PA (n = 15)	Streptokinase (n = 15)	Anistreplase (n = 13)	rt-PA (n = 15)	Streptokinase (n = 15)	Anistreplase (n = 13)
Baseline	1.26	1.11	1.08	6.4	6.2	8.6	2.8	2.7	4.1
	(0.79–2.06)	(0.71–1.44)	(0.7–1.88)	(5.6–9)	(4.5–8.8)	(6–17.8)	(1.6–4.1)	(0.9–2.8)	(1.3–8.8)
90 min	3.13*	2.57*	2.99*	16.8*	16.4*	24.5*	2.5	2.5	4.5
	(2.42–3.95)	(2.06–3.13)	(1.6–4.18)	(13.7–20.2)	(10.9–20.5)	(15.8–33.5)	(1.4–4.2)	(1.4–4)	(2–6)
24 h	0.95	1.05	1.03	4.2	10.8	10	1.3†	1.85	2.3
	(0.83–1.82)	(0.59–1.41)	(0.65–1.12)	(3.5–6)	(5.8–17.4)	(3.4–14.9)	(0.8–1.7)	(1.1–3)	(1.5–5)
48 h	1.06	0.94	1.28	5.7	4.8	7.6	1.3†	1.1†	2.4†
	(0.77–1.82)	(0.81–1.23)	(1.01–1.84)	(3.5–7.7)	(4.1–6.5)	(4–16.7)	(1.0–2.2)	(0.8–2.2)	(2.3–3.1)

*$p < 0.01$ versus baseline and 24 and 48 h. †$p < 0.05$ versus baseline. Data presented are median values (25th to 75th percentile).

2.7) in the streptokinase group and −1.2 μg/liter (interquartile range −3.55 to 0.35) in the anistreplase group (p = 0.57).

No differences were detected among the three groups in terms of median changes in fibrinopeptide A at different time points. At 90 min there was a median change of 1.1 nmol/liter (interquartile range 0 to 3.8) in the rt-PA group, 0.9 nmol/liter (interquartile range −0.35 to 2.5) in the streptokinase group and 0.8 nmol/liter (interquartile range −4.8 to 1.3) in the anistreplase group (p = 0.25). Compared with baseline, at 24 h there was a median change of −1.35 nmol/liter (interquartile range −2 to −0.1) in the rt-PA group, 0 nmol/liter (interquartile range −1.05 to 0.35) in the streptokinase group and 0.1 nmol/liter (interquartile range −2.55 to 0.85) in the anistreplase group (p = 0.42). At 48 h there was a median change of −0.7 nmol/liter (interquartile range −1 to 0.1) in the rt-PA group, −0.2 nmol/liter (interquartile range −1.9 to 0.9) in the streptokinase group and −1.2 nmol/liter (interquartile range −3.55 to 0.35) in the anistreplase group (p = 0.66).

Discussion

Thrombin generation and activity during thrombolysis. In the present study the plasma levels of prothrombin fragment 1 + 2 and thrombin–antithrombin complex (indexes of thrombin generation by factor Xa activity) and fibrinopeptide A (an index of thrombin activity on fibrinogen) were measured at different times in patients with myocardial infarction receiving different thrombolytic drugs in conjunction with heparin. The data show that during thrombolytic therapy and concomitant intravenous heparin, thrombin activity is efficiently inhibited, but there is an increase in thrombin generation. Thrombin generation increased 90 min after thrombolytic therapy and was similar to baseline after 24 and 48 h, whereas thrombin activity showed no change at 90 min and decreased after 24 and 48 h. Previous studies have shown that there is an increase in thrombin activity in patients receiving thrombolytic therapy (3–8) that does not occur when heparin is given before and during treatment (6). In a pilot study (19), we also observed a marked increase in both thrombin generation and activity in patients receiving thrombolytic therapy with rt-PA or streptokinase in the absence of concomitant anticoagulation with heparin. Therefore, we designed the present study to evaluate the behavior of coagulation system markers in patients receiving thrombolytic therapy and concomitant heparin treatment.

The data show that there is no significant increase in fibrinopeptide A in patients receiving heparin before thrombolytic therapy. Thus, despite experimental evidence that clot-bound thrombin is protected from inactivation by antithrombin III–dependent anticoagulants (20), in vivo biochemical data seem to suggest that during thrombolytic therapy, the contribution of clot-bound thrombin to the increase in fibrinopeptide A plasma levels is minor and that intravenous heparin is effective in preventing an increase in thrombin activity in most patients. A previous study (5) has shown that thrombin generation is increased during rt-PA infusion in patients not receiving heparin. Our data show that heparin is not effective in inhibiting an increase in thrombin generation in patients receiving thrombolytic therapy. An in vitro study (17) on human whole blood has shown that factor Xa binding to activated platelets leads to protection against inactivation by the heparin–antithrombin III complex, and the degree of protection is closely related to the extent of prothrombin activation.

It is possible to speculate that at high rates of in vivo thrombin generation, as observed in patients with acute myocardial infarction, a significant amount of factor Xa is bound to activated platelets, thus allowing this serine protease to resist the action of heparin. In addition, the direct activation of platelets (1,2) and factor Va (21) during thrombolysis, as well as the reexposure of the ruptured plaque to blood flow, might lead to the generation of more factor Xa and, consequently, to thrombin generation. The increase in prothrombin fragment 1 + 2 and thrombin–antithrombin complex plasma levels in the first hours after the initiation of thrombolysis despite heparin treatment further supports this hypothesis.

Thrombin generation and activity according to the thrombolytic drug administered. The changes in the measured coagulation activation markers were similar regardless of the thrombolytic drug used. Although previous in vitro studies (22,23) have suggested that the effects of fibrin-specific or

non–fibrin-specific thrombolytic agents on the activation of hemostatic mechanisms may be different, clinical studies have found a similar early increase in coagulation activation markers. Owen et al. (4) have shown that patients receiving heparin in association with streptokinase or rt-PA presented a similar increase in plasma fibrinopeptide A levels and concluded that this transient elevation was a result of transient thrombin activity and that the close similarity in the observed increases in patients receiving streptokinase or rt-PA indicates that this relates more to the thrombolysis itself than to the individual thrombolytic agent. Gulba et al. (24), who prospectively studied thrombin–antithrombin complex plasma levels in patients who received rt-PA or pro-urokinase after heparin treatment and then underwent angiography 90 min and 24 to 36 h after thrombolytic therapy, did not detect any significant differences between the effects of the two thrombolytic regimens.

Limitations of the study. One possible limitation of the present study concerns the use of plasma fibrinopeptide A levels as a marker of local coronary thrombin activity. Only a percent of patients with acute myocardial infarction show elevated fibrinopeptide A levels. Furthermore, elevated levels have been found in patients with both thrombotic and inflammatory disorders of various types, thereby limiting the diagnostic specificity of the assay. However, our study was not aimed at assessing a link with coronary thrombosis or its evolution and coagulation activation markers but at monitoring the time course of thrombin generation and activity during thrombolysis and concomitant heparin administration.

Another possible limitation concerns the high degree of variability of the coagulation system markers, which limits the power of the study to detect significant increases in fibrinopeptide A during thrombolysis or different behaviors among three thrombolytic regimens. The primary aim of the study was to assess hemostatic system activation in patients with acute myocardial infarction receiving thrombolytic therapy in conjunction with heparin and not to make a direct comparison of specific treatments. However, with an alpha error of 0.05 and a beta of 0.1, our sample size allows the detection of a difference between the most and the least activating agents of 18% for prothrombin fragment 1 + 2, 43% for thrombin-antithrombin complex and 40% for fibrinopeptide A.

Conclusions. The finding of increased thrombin generation during thrombolysis despite the concomitant administration of heparin raises a number of questions. Apart from catalyzing the conversion of fibrinogen to fibrin, thrombin is a potent activator of a variety of cell events, such as platelet aggregation (25), secretion and formation of thromboxane A_2 (26), contraction of smooth muscle cells (27,28) and expression of tissue factor on endothelial cells (29), all of which may have detrimental effects at the site of highly unstable postthrombolysis residual lesions (30,31). It is tempting to speculate that the remarkably favorable effects of direct antithrombin agents compared with heparin (32) during thrombolysis may be attributable to a more extensive inhibition of blood coagulation, which includes both fibrin formation and platelet aggregation. In the setting of coronary thrombolysis, thrombin is the main factor responsible for platelet activation; therefore, the favorable results obtained with aspirin (10), which only inhibits the thromboxane-induced platelet aggregation, might be further improved by direct thrombin inhibition.

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